

Original Research Article

A STUDY ON EVALUATION OF Bcl-2 AND Ki67 LEVELS IN GASTRIC ADENOCARCINOMA

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ABSTRACT

Background: Gastric cancer is one of the leading causes of cancer related deaths. Immunoexpression of Bcl-2 and Ki67 levels are used to assess the prognosis and aggressiveness of tumor.

Materials and Methods: A total of 50 cases with gastric cancer who were operated in Mallareddy medical college for women over a period of 2 years were included. The immunoexpression levels of Bcl-2 and Ki67 were assessed.

Results: Bcl-2 and Ki67 levels were higher in higher grades of gastric adenocarcinoma.

Conclusion: Bcl-2 and Ki67 owing to its anti-apoptotic and pro-proliferative action respectively, are indicators of tumor aggressiveness.

Keywords: Bcl-2, Ki67, Gastric adenocarcinoma, Immunotyping.

INTRODUCTION

Gastric cancer is the fifth most common cancer and the fourth most common cause of cancer-related deaths worldwide.^[1] In recent years the incidence of gastric cancer is on rise despite of progress in the diagnostic and therapeutic modalities.^[2] Several factors contribute to the development of gastric cancer, such as H. pylori infection, dietary habits (such as consumption of smoked or pickled foods), tobacco smoking, alcohol consumption, obesity, genetic predisposition, and certain medical conditions like chronic gastritis or gastric ulcers.^[3]

The development of gastric cancer involves a complex interplay of genetic, environmental, and lifestyle factors. Chronic inflammation, particularly associated with H. pylori infection, plays a crucial role in gastric carcinogenesis.^[4]

Gastric cancer remains a significant public health challenge globally, and ongoing research efforts are focused on improving early detection methods, understanding molecular mechanisms underlying the disease, and developing more effective treatment strategies to combat this malignancy.

The B-cell lymphoma 2 (Bcl-2) gene are members of the Bcl-2 family, which includes both pro-apoptotic (e.g., Bax, Bak) and anti-apoptotic (e.g., Bcl-2, Bcl-xL) members. Bcl-2 proteins regulate apoptosis by controlling mitochondrial outer membrane

permeability and release of apoptotic factors, thereby influencing cell survival or death.^[5,6]

Overexpression of Bcl-2 in gastric cancer cells causes resistance to apoptosis by inhibiting the intrinsic apoptotic pathway, thereby promoting cell survival and tumor progression.^[7]

Ki-67 is a tumor marker used to measure cell proliferation and assess tumor aggressiveness in various cancers, including gastric cancer. Ki-67 labeling index (percentage of positively stained tumor cells) is commonly used to quantify cell proliferation activity and predict patient outcomes in gastric cancer.^[8]

This study was conducted with an aim to evaluate the immunoexpression of Bcl-2 and Ki67 tumor markers in patients with gastric adenocarcinoma.

MATERIALS AND METHODS

This prospective study was conducted in the Department of Pathology, Mallareddy Medical college for women over a period of two years. All patients with gastric cancer, who had adenocarcinoma type on histopathological examination and who were operated in the hospital were included. Patients with other types of gastric carcinoma and patients who were operated outside the place of study were excluded from the study. A total of 50 patients were selected for the study.

A detailed history and general examination of all patients was done. A study protocol was made which was approved by the institutional ethical committee. A written informed consent was obtained from all participants.

The tissue specimens were fixed using 10% formalin and paraffin embedded using standard technique. Each tissue specimen was microtomed into 4 sections for staining. Two sections were stained with H& E stain, one with Bcl-2 and other for Ki67 stain.

The levels of Bcl-2 expression were scored according to the fraction of stained tumor cells. The Bcl-2 score was evaluated as negative (0) when no positive cell were observed; 1+ when up to 30% of the tumor cells were positive; 2+ when 31%–70% of the tumor cells were positive and 3+ when 71–100% of tumor cells were positive

Levels of Ki67 expression were scored according to the proliferative index which is as follows: no cells positive = 0; 1+ = when ≤1% of cells are positive; 2+ = when >1-5% of cells are positive; 3+ = when >5–10% of cells are positive; 4+ =when >10–20% of cells are positive and 5+ = when >20% of cells are positive.

Statistical Analysis

All analyses were performed using the SPSS software. Quantitative variables were compared using the Pearson's chi-square test. P value was said to be significant if it was <0.05.

RESULTS

A total of 50 patients with gastric cancer were included. Amongst the 50 patients, there were 30 males and 20 females. The mean age of study population was 56.4 years with range being from 47 years to 85 years.

All tissues were subjected to bcl-2 staining and Ki67 staining. Bcl-2 staining was localized to the cytoplasm and Ki67 was localized to the nuclei of the gastric cancer cells.

Immunostaining for Bcl-2

Amongst all the 50 cases with gastric cancer, Bcl-2 positivity was seen in 42 cases (84%), while the rest 8 cases tested negative, i.e. 0% of the cells were negative (16%). Majority of the cells were 3+ and above, indicating the advanced stage of the carcinoma.

Table 1: Expression of Bcl-2

Score	No of cases	Grade 1	Grade 2	Grade 3	Grade 4	Percentage
0	8	6	1	1	0	16%
1+	10	4	2	4	0	20%
2+	15	0	3	6	6	30%
3+	17	0	1	7	9	34%
Total	50	10	7	18	15	100%

P value = 0.01713 (significant as p value is <0.05)

Immunostaining for Ki67: Amongst all the 50 cases with gastric adenocarcinoma, positivity for Ki67 was seen in 90% (n=45) of the cases. Amongst the ones who tested positive, majority of them had Ki67 scores above 10% (scores of 4+ and above).

Table 2: Expression of Ki67

Score	No of cases	Grade 1	Grade 2	Grade 3	Grade 4	Percentage
0	5	3	1	1	0	10%
1+	4	1	3	0	0	8%
2+	8	2	4	2	0	16%
3+	9	0	1	3	5	18%
4+	14	0	0	6	8	28%
5+	10	0	0	4	6	20%
Total	50	6	9	16	19	100%

P value = 0.04179 (significant as p value is <0.05)

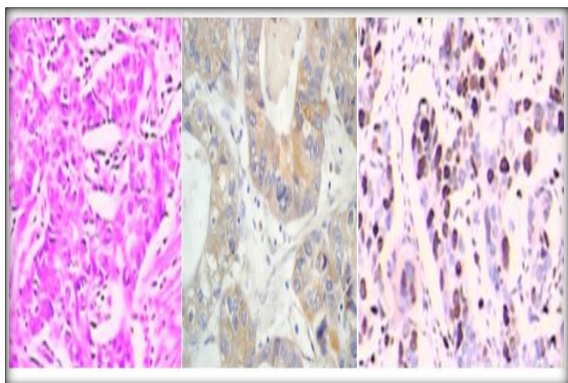


Figure 1: Figure 1-3: Histopathological images of gastric cancer; Figure 1- H&E staining; Figure 2- Bcl staining; Figure 3- Ki67 staining

DISCUSSIONS

Bcl-2 is an anti-apoptotic tumor markers belonging to the Bcl-2 group of proteins which are localized in the membranes, especially the mitochondrial membrane, endoplasmic reticulum. Dysregulation of Bcl-2 expression or function has been implicated in the pathogenesis of various cancers including lymphoma, leukemia, breast cancer, prostate cancer, and gastric adenocarcinoma.^[9-11]

Ki-67 is a non-histone nuclear protein antigen encoded by the MKI67 gene. It plays a critical role in the regulation of cell proliferation by being present during all active phases of the cell cycle (G1, S, G2, and mitosis), but absent in resting (G0) cells. Ki-67 expression is commonly assessed by immunohistochemistry (IHC) in tumor tissues. High Ki-67 expression levels indicate increased cell proliferation and are associated with more aggressive tumor behavior, higher tumor grade, and poorer prognosis in various cancer types.^[8,12,13]

In present study, majority of the cells show bcl-2 positivity with most of the cells having positivity of 3+. Grade 4 of cancer which indicates highly aggressive cellular proliferation has the the highest level of bcl-2 and Ki67 immunoexpression levels. This is suggestive of the highly proliferative nature of the tumor.

In present study, 84% of the patients have Bcl-2 expression. This is higher when compared to study done by Mariusz et al¹⁴ and Tsamandas et al¹⁵ who observed 55.8% and 67% positivity respectively.

In present study, levels of Bcl-2 expression has significant correlation with the tumor grade. However, Martin-Arruti et al reported that Bcl-2 expression has no significant correlation.^[16]

In present study, 90% of the cases have Ki67 expression, with majority of the levels being significantly higher with higher grades of cancer. Ko et al,^[17] conducted a study on 320 patients with gastric cancer to investigate the prognostic importance of Ki67 expression in patients with early gastric cancer. They concluded the study by saying that high levels of Ki67 are suggestive of poor prognosis.

CONCLUSION

This study concludes that Bcl-2 and Ki67 are markers of highly proliferative tumours. The routine evaluation of Ki67 and Bcl-2 levels could be a useful tool in identification of patient with more aggressive disease and contribute to a better the rapetic approach

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